CLAIMS

What is claimed is:

1. Method of simultaneous screening for one or more gene insertion mutants in a population of any organism or cell line derived thereof by:

a) preparing an insertion element mutant library originating from a defined population of an organism or cell line wherein said insertion(s) have to be detected,

b) amplifying the insertion element flanking sequences from said insertion element mutant library,

c1) either fixing the set of thus obtained nucleic acid amplification products representing said insertion element flanking sequences derived from said insertion element mutant library to a solid support as target for hybridization, or

c2) producing a set of labelled amplification products representing said insertion element flanking sequences derived from said insertion element mutant library to use as probe to hybridize to a solid support to which one or more nucleic acids have been fixed as target(s) for hybridisation.

2. Method according to claim 1 wherein the thus obtained nucleic acid amplification products in step b) are obtained by iPCR using at least one primer or a set of primers based on the sequence of the insertion element.

3. Method according to claim 2 wherein the iPCR is performed by

a) digesting the nucleic acid sequences of said insertion element mutant library with a restriction enzyme which optionally recognizes motifs of four nucleotides in the genomic DNA, or with a combination of restriction enzymes resulting in a collection of amplifyable fragments,

b) self ligation of the genomic fragments thus obtained and either

c1) amplification of insertion element flanking sequences using a set of internal primers or

c2) amplification of insertion element flanking sequences using a (set of) primers based on the terminal part of the insertion element.

4. Method according to claim 3 wherein the amplification products of step c1 are re-amplified using at least one primer or a set of two nested primers based on the sequence of the insertion element.

5. Method according to claim 1 wherein the amplification products in step b) are obtained by transposon display amplification.

6. Method according to claim 5 wherein transposon display amplification is performed by

a) digesting the nucleic acid sequences of said insertion element mutant library with a first restriction enzyme that recognizes six conserved nucleotides in the insertion element and with a second restriction enzyme that recognizes a motif of four nucleotides in the genome generating at least one restriction fragment per insertion containing at least the hexacutter site, a part of the insertion element, and part of the insertion element flanking sequence,

b) ligation of a biotinylated adaptor to the hexacutter sites and a ligation of a second adaptor to the tetracutter sites of the restriction fragments generated in a),

c) selection of biotinylated restriction fragments using magnetic streptavidin beads,

d) amplification of insertion element flanking sequences using a primer based on the sequence of the biotinylated adaptor and on the insertion element sequence and a primer complementary to the second adaptor,

e) re-amplification of said insertion element flanking sequences using a nested primer based on the insertion element and a primer complementary to the second adaptor.

7. Method according to any of the preceeding claims wherein the solid support is a filter, micro-array or chip containing nucleic acid sequences.

8. Method according to any of the preceeding claims wherein the nucleic acid sequence is genomic DNA or cDNA.

9. Method according to any of the preceeding claims werein the insertion element mutant library comprises of 30 DNA samples from 100 plants each.

10. Method according to claim 9 wherein the insertion element mutant library is built in a 3D array of 10 Block, 10 Row and 10 Column pool each containing DNA of 100 plants characterised by the three coordinates B, R, C.

11. Method according to any of the claims 1-4 wherein BfaI is used as restriction enzyme.

12. Method according to the claims 5 or 6 wherein MseI and/or MunI are used as restriction enzyme.

13. Kit for performing any of the methods of claim 1-12 comprising at least DNA samples of an insertion element mutant library and optionally a set of restriction enzymes and/or primers.

14. Kit for performing any of the methods of claim 1-12 comprising at least a set of amplified insertion element flanking sequences.

15. Kit according to claim 14 wherein the set of insertion element flanking sequences have been fixed on a solid support such as a filter, micro-array or microchip containing nucleic acid sequences.

16. Kit according to claims 14 or 15 wherein the set of insertion element flanking sequences is either present in soluble form or dried form.

17. Kit according to claim 16 wherein the set of insertion element flanking sequences are labelled with for instance fluorescein.